

CLAIMS

What is claimed is:

1. An isolated polynucleotide comprising a first nucleotide sequence encoding a polypeptide of at least 160 amino acids that has at least 80% identity based on the Clustal method of alignment when compared to a polypeptide selected from the group consisting of SEQ ID NOs:2, 4, 6, 8, 10, 22, 24, 26, 28, and 30, or a second nucleotide sequence comprising the complement of the first nucleotide sequence.
2. An isolated polynucleotide comprising a first nucleotide sequence encoding a polypeptide of at least 150 amino acids that has at least 95% identity based on the Clustal method of alignment when compared to a polypeptide selected from the group consisting of SEQ ID NOs:12, 14, 16, 18, 20, 32, 34, 36, and 38, or a second nucleotide sequence comprising the complement of the first nucleotide sequence.
3. The isolated polynucleotide of Claim 1 or Claim 2, wherein the first nucleotide sequence consists of a nucleic acid sequence selected from the group consisting of SEQ ID NOs:1, 3, 5, 7, 9, 11, 13, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 35, and 37 that codes for the polypeptide selected from the group consisting of SEQ ID NOs:2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, and 38.
4. The isolated polynucleotide of Claim 1 or Claim 2 wherein the nucleotide sequences are DNA.
5. The isolated polynucleotide of Claim 1 or Claim 2 wherein the nucleotide sequences are RNA.
6. A chimeric gene comprising the isolated polynucleotide of Claim 1 or Claim 2 operably linked to suitable regulatory sequences.
7. An isolated host cell comprising the chimeric gene of Claim 6.
8. A host cell comprising an isolated polynucleotide of Claim 1 or Claim 2.
9. The host cell of Claim 8 wherein the host cell is selected from the group consisting of yeast, bacteria, plant, and virus.
10. A virus comprising the isolated polynucleotide of Claim 1 or Claim 2.
11. A polypeptide of at least 160 amino acids that has at least 80% identity based on the Clustal method of alignment when compared to a polypeptide selected from the group consisting of SEQ ID NOs:2, 4, 6, 8, 10, 22, 24, 26, 28, and 30.
12. A polypeptide of at least 150 amino acids that has at least 95% identity based on the Clustal method of alignment when compared to a polypeptide selected from the group consisting of SEQ ID NOs:12, 14, 16, 18, 20, 32, 34, 36, and 38.
13. A method of selecting an isolated polynucleotide that affects the level of expression of a vitamin E biosynthetic enzyme polypeptide in a plant cell, the method comprising the steps of:

(a) constructing an isolated polynucleotide comprising a nucleotide sequence of at least one of 30 contiguous nucleotides derived from an isolated polynucleotide of Claim 1 or Claim 2;

(b) introducing the isolated polynucleotide into a plant cell; and

5 (c) measuring the level of a polypeptide in the plant cell containing the polynucleotide to provide a positive selection means.

14. The method of Claim 13 wherein the isolated polynucleotide consists of a nucleotide sequence selected from the group consisting of SEQ ID NOs:1, 3, 5, 7, 9, 11, 13, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 35, and 37 that codes for the polypeptide selected from
10 the group consisting of SEQ ID NOs:2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, and 38.

15. A method of selecting an isolated polynucleotide that affects the level of expression of a vitamin E biosynthetic enzyme polypeptide in a plant cell, the method comprising the steps of:

15 (a) constructing an isolated polynucleotide of Claim 1 or Claim 2;

(b) introducing the isolated polynucleotide into a plant cell; and

(c) measuring the level of polypeptide in the plant cell containing the polynucleotide to provide a positive selection means.

16. A method of obtaining a nucleic acid fragment encoding a vitamin E
20 biosynthetic enzyme polypeptide comprising the steps of:

(a) synthesizing an oligonucleotide primer comprising a nucleotide sequence of at least one of 30 contiguous nucleotides derived from a nucleotide sequence selected from the group consisting of SEQ ID NOs:1, 3, 5, 7, 9, 11, 13, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 35, 37, and the complement of such nucleotide sequences; and

25 (b) amplifying a nucleic acid sequence using the oligonucleotide primer.

17. A method of obtaining a nucleic acid fragment encoding a vitamin E biosynthetic enzyme polypeptide comprising the steps of:

(a) probing a cDNA or genomic library with an isolated polynucleotide comprising at least one of 30 contiguous nucleotides derived from a nucleotide sequence selected from the group consisting of SEQ ID NOs:1, 3, 5, 7, 9, 11, 13, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 35, 37 and the complement of such nucleotide sequences;

(b) identifying a DNA clone that hybridizes with the isolated polynucleotide;

(c) isolating the identified DNA clone; and

35 (d) sequencing the cDNA or genomic fragment that comprises the isolated DNA clone.

18. A method for evaluating at least one compound for its ability to inhibit the activity of a vitamin E biosynthetic enzyme, the method comprising the steps of:

(a) transforming a host cell with a chimeric gene comprising a nucleic acid fragment encoding a vitamin E biosynthetic enzyme, operably linked to suitable regulatory sequences;

5 (b) growing the transformed host cell under conditions that are suitable for expression of the chimeric gene wherein expression of the chimeric gene results in production of the vitamin E biosynthetic enzyme encoded by the operably linked nucleic acid fragment in the transformed host cell;

(c) optionally purifying the vitamin E biosynthetic enzyme expressed by the transformed host cell;

10 (d) treating the vitamin E biosynthetic enzyme with a compound to be tested; and

(e) determining the activity of the vitamin E biosynthetic enzyme that has been treated with the compound, thereby selecting compounds with potential for inhibitory activity.

15 19. A composition comprising the isolated polynucleotide of Claim 1 or Claim 2.

20. A composition comprising the isolated polypeptide of Claim 11.

21. An isolated polynucleotide comprising the nucleotide sequence having at least one of 30 contiguous nucleotides derived from a nucleic acid sequence selected from the group consisting of SEQ ID NOs:1, 3, 5, 7, 9, 11, 13, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 20 35, 37 and the complement of such sequences.

22. An expression cassette comprising an isolated polynucleotide of Claim 1 or Claim 2 operably linked to a promoter.

23. A method for positive selection of a transformed cell comprising:

25 (a) transforming a host cell with an expression cassette of Claim 22; and

(b) growing the transformed host cell under conditions which allow expression of the polynucleotide in an amount sufficient to complement a null mutant to provide a positive selection means.

24. The method of Claim 23 wherein the plant cell is a monocot.

25. The method of Claim 23 wherein the plant cell is a dicot.

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